OIPE

RAW SEQUENCE LISTING DATE: 08/15/2001
PATENT APPLICATION: **US/09/876,348A** TIME: 14:16:16

Input Set : **A:\Rb125seq.txt**
Output Set: **N:\CRF3\08152001\I876348A.raw**

ENTERED

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  3 <110> APPLICANT: Horwath, K. L. and Myers, K. L.
  5 <120> TITLE OF INVENTION: Nucleic Acid Sequences Encoding Type III Tenebrio
  6       Antifreeze Proteins and Method for Assaying Activity.
  8 <130> FILE REFERENCE: RB-125-RI
 10 <140> CURRENT APPLICATION NUMBER: 09/876,348A
C--> 11 <141> CURRENT FILING DATE: 2001-08-09
 13 <150> PRIOR APPLICATION NUMBER: 60/210,446
 14 <151> PRIOR FILING DATE: 2000-06-08
 16 <160> NUMBER OF SEQ ID NOS: 48
 18 <170> SOFTWARE: Microsoft Word
 20 <210> SEQ ID NO: 1
 21 <211> LENGTH: 19
 22 <212> TYPE: PRT
 23 <213> ORGANISM: Tenebrio molitor
 25 <223> OTHER INFORMATION: N-terminal sequence of protein Tm 12.86
 27 <400> SEQUENCE: 1
 28 Leu Thr Asp Glu Gln Ile Gln Lys Arg Asn Lys Ile Ser Lys Glu Cys
 29 1               5                  10                  15
 31 Gln Gln Val
 32         19
 34 <210> SEQ ID NO: 2
 35 <211> LENGTH: 576
 36 <212> TYPE: DNA
 37 <213> ORGANISM: Tenebrio molitor
 39 <223> OTHER INFORMATION: Non-his-tagged, signal plus, Tm 13.17
 41 <400> SEQUENCE: 2
 42 gtggatccaa agaattcggc acgagactac taag atg aag ttg ctc            46
 43                                     Met Lys Leu Leu
 44                                                -15
 46 tgt tgt cta atc tcc ctc att ctg ttg gtc aca gtt cag gcc ctg      91
 47 Cys Cys Leu Ile Ser Leu Ile Leu Leu Val Thr Val Gln Ala Leu
 48                 -10                 -5                  1
 50 acc gag gca caa att gag aaa ctg aac aag atc agc aaa aaa tgt     136
 51 Thr Glu Ala Gln Ile Glu Lys Leu Asn Lys Ile Ser Lys Lys Cys
 52             5                  10                  15
 54 caa aat gaa agt gga gtg tcg caa gag atc ata acc aaa gct cgc     181
 55 Gln Asn Glu Ser Gly Val Ser Gln Glu Ile Ile Thr Lys Ala Arg
 56             20                  25                  30
 58 aac ggt gac tgg gag gac gat cct aaa ctg aaa cgc caa gtt ttt     226
 59 Asn Gly Asp Trp Glu Asp Asp Pro Lys Leu Lys Arg Gln Val Phe
 60             35                  40                  45
 62 tgc gtg gcc agg aac gcc ggt ctg gcc acg gaa tcg gga gag gtg     271
 63 Cys Val Ala Arg Asn Ala Gly Leu Ala Thr Glu Ser Gly Glu Val
 64             50                  55                  60
 66 gtg gtc gac gtg ttg agg gag aag gtg agg aag gtc act gac aac     316
 67 Val Val Asp Val Leu Arg Glu Lys Val Arg Lys Val Thr Asp Asn
 68             65                  70                  75
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RAW SEQUENCE LISTING DATE: 08/15/2001
PATENT APPLICATION: US/09/876,348A TIME: 14:16:16

Input Set : A:\Rbl25seq.txt
Output Set: N:\CRF3\08152001\I876348A.raw

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70 gac gaa gaa act gag aaa atc atc aat aag tgc gcc gtc aag aga     361
71 Asp Glu Glu Thr Glu Lys Ile Ile Asn Lys Cys Ala Val Lys Arg
72             80                  85                  90
74 gat act gtt gaa gag acg gtg ttc aat act ttc aaa tgt gtc atg     406
75 Asp Thr Val Glu Glu Thr Val Phe Asn Thr Phe Lys Cys Val Met
76             95                 100                 105
78 aaa aac aag cca aag ttc tca cca gtt gat tga accaccacga         449
79 Lys Asn Lys Pro Lys Phe Ser Pro Val Asp
80             110                 115
82 ctagtagatg gttcaaatgg tgtgctttac atataaaaat aaagtgtttc         499
84 tgatgtaaaa aaaaaaaaaa aaaaaaaaaa aactcgagag tattctagag         549
86 cggccgcggg cccatcgttt tccaccc                                 576
89 <210> SEQ ID NO: 3
90 <211> LENGTH: 134
91 <212> TYPE: PRT
92 <213> ORGANISM: Tenebrio molitor
94 <223> OTHER INFORMATION: Precursor Protein for Tm 13.17
96 <400> SEQUENCE: 3
97 Met Lys Leu Leu Cys Cys Leu Ile Ser Leu Ile Leu Leu Val Thr Val
98             -15                 -10                 -5
100 Gln Ala Leu Thr Glu Ala Gln Ile Glu Lys Leu Asn Lys Ile Ser Lys
101         1               5                  10
103 Lys Cys Gln Asn Glu Ser Gly Val Ser Gln Glu Ile Ile Thr Lys Ala
104 15                  20                  25                  30
106 Arg Asn Gly Asp Trp Glu Asp Asp Pro Lys Leu Lys Arg Gln Val Phe
107                 35                  40                  45
109 Cys Val Ala Arg Asn Ala Gly Leu Ala Thr Glu Ser Gly Glu Val Val
110             50                  55                  60
112 Val Asp Val Leu Arg Glu Lys Val Arg Lys Val Thr Asp Asn Asp Glu
113         65                  70                  75
115 Glu Thr Glu Lys Ile Ile Asn Lys Cys Ala Val Lys Arg Asp Thr Val
116     80                  85                  90
118 Glu Glu Thr Val Phe Asn Thr Phe Lys Cys Val Met Lys Asn Lys Pro
119 95                  100                 105                 110
121 Lys Phe Ser Pro Val Asp
122                 115
125 <210> SEQ ID NO: 4
126 <211> LENGTH: 116
127 <212> TYPE: PRT
128 <213> ORGANISM: Tenebrio molitor
130 <223> OTHER INFORMATION: Mature Protein for Tm 13.17
132 <400> SEQUENCE: 4
133 Leu Thr Glu Ala Gln Ile Glu Lys Leu Asn Lys Ile Ser Lys Lys Cys
134 1               5                  10                  15
136 Gln Asn Glu Ser Gly Val Ser Gln Glu Ile Ile Thr Lys Ala Arg Asn
137             20                  25                  30
139 Gly Asp Trp Glu Asp Asp Pro Lys Leu Lys Arg Gln Val Phe Cys Val
140         35                  40                  45
142 Ala Arg Asn Ala Gly Leu Ala Thr Glu Ser Gly Glu Val Val Val Asp
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RAW SEQUENCE LISTING DATE: 08/15/2001
PATENT APPLICATION: US/09/876,348A TIME: 14:16:16

Input Set : A:\Rbl25seq.txt
Output Set: N:\CRF3\08152001\I876348A.raw

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143     50                  55                  60
145 Val Leu Arg Glu Lys Val Arg Lys Val Thr Asp Asn Asp Glu Glu Thr
146 65                  70                  75                  80
148 Glu Lys Ile Ile Asn Lys Cys Ala Val Lys Arg Asp Thr Val Glu Glu
149                 85                  90                  95
151 Thr Val Phe Asn Thr Phe Lys Cys Val Met Lys Asn Lys Pro Lys Phe
152             100                 105                 110
154 Ser Pro Val Asp
155         115
158 <210> SEQ ID NO: 5
159 <211> LENGTH: 481
160 <212> TYPE: DNA
161 <213> ORGANISM: Tenebrio molitor
163 <223> OTHER INFORMATION: Non-His-tagged, Signal plus, Clone 2.2
165 <400> SEQUENCE: 5
166 ggcacgagca aaa atg aaa ctc ctc ttg tgc ttt gcg ttc gcc gcc     46
167                 Met Lys Leu Leu Leu Cys Phe Ala Phe Ala Ala
168                             -15                 -10
170 atc gtc atc gga gct cag gct ctc acc gac gaa cag ata cag aaa     91
171 Ile Val Ile Gly Ala Gln Ala Leu Thr Asp Glu Gln Ile Gln Lys
172         -5                  1                   5
174 agg aac aag atc agc aaa gaa tgc cag cag gtg tcc gga gtg tcc   136
175 Arg Asn Lys Ile Ser Lys Glu Cys Gln Gln Val Ser Gly Val Ser
176     10                  15                  20
178 caa gag acg atc gac aaa gtc cgc aca ggt gtc ttg gtc gat gat   181
179 Gln Glu Thr Ile Asp Lys Val Arg Thr Gly Val Leu Val Asp Asp
180     25                  30                  35
182 ccc aaa atg aag aag cac gtc ctc tgc ttc tcg aag aaa act gga   226
183 Pro Lys Met Lys Lys His Val Leu Cys Phe Ser Lys Lys Thr Gly
184     40                  45                  50
186 gtg gca acc gaa gcc gga gac acc aat gtg gag gta ctc aaa gcc   271
187 Val Ala Thr Glu Ala Gly Asp Thr Asn Val Glu Val Leu Lys Ala
188     55                  60                  65
190 aag ctg aag cat gtg gcc agc gac gaa gag gtg gac aag atc gtg   316
191 Lys Leu Lys His Val Ala Ser Asp Glu Glu Val Asp Lys Ile Val
192     70                  75                  80
194 cag aag tgc gtg gtc aag aag gcc aca cca gag gaa acg gct tat   361
195 Gln Lys Cys Val Val Lys Lys Ala Thr Pro Glu Glu Thr Ala Tyr
196     85                  90                  95
198 gac acc ttc aag tgt att tac gac agt aaa cct gat ttc tct cct   406
199 Asp Thr Phe Lys Cys Ile Tyr Asp Ser Lys Pro Asp Phe Ser Pro
200     100                 105                 110
202 att gat taa ttgttttgta tttgactgaa ttttgacaat aaaggtaata       455
203 Ile Asp
204     115
206 tcgttatgta aaaaaaaaaa aaaaaa                                 481
209 <210> SEQ ID NO: 6
210 <211> LENGTH: 482
211 <212> TYPE: DNA
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RAW SEQUENCE LISTING DATE: 08/15/2001
PATENT APPLICATION: US/09/876,348A TIME: 14:16:16

Input Set : A:\Rb125seq.txt
Output Set: N:\CRF3\08152001\I876348A.raw

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212 <213> ORGANISM: Tenebrio molitor
214 <223> OTHER INFORMATION: Non-His-tagged, Signal plus, Clone 2.3
216 <400> SEQUENCE: 6
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218                Met Lys Leu Leu Leu Cys Phe Ala Phe Ala Ala
219                            -15                 -10
221 atc gtc atc gga gct cag gct ctc acc gac gaa cag ata cag aaa    91
222 Ile Val Ile Gly Ala Gln Ala Leu Thr Asp Glu Gln Ile Gln Lys
223         -5                  1                   5
225 agg aac aag atc agc aaa gaa tgc cag cag gtg tcc gga gtg tcc   136
226 Arg Asn Lys Ile Ser Lys Glu Cys Gln Gln Val Ser Gly Val Ser
227     10                  15                  20
229 caa gag acg atc gac aaa gtc cgc aca ggt gtc ttg gtc gac gat   181
230 Gln Glu Thr Ile Asp Lys Val Arg Thr Gly Val Leu Val Asp Asp
231     25                  30                  35
233 ccc aaa atg aag aag cac gtc ctc tgc ttc tcg aag aaa act gga   226
234 Pro Lys Met Lys Lys His Val Leu Cys Phe Ser Lys Lys Thr Gly
235     40                  45                  50
237 gtg gca acc gaa gcc gga gac acc aat gtg gag gta ctc aaa gcc   271
238 Val Ala Thr Glu Ala Gly Asp Thr Asn Val Glu Val Leu Lys Ala
239     55                  60                  65
241 aag ctg aag cat gtg gcc agc gac gaa gaa gtg gac aag atc gtg   316
242 Lys Leu Lys His Val Ala Ser Asp Glu Glu Val Asp Lys Ile Val
243     70                  75                  80
245 cag aag tgc gtg gtc aag aag gcc aca cca gag gaa acg gct tat   361
246 Gln Lys Cys Val Val Lys Lys Ala Thr Pro Glu Glu Thr Ala Tyr
247     85                  90                  95
249 gac acc ttc aag tgt att tac gac agt aaa cct gat ttc tct cct   406
250 Asp Thr Phe Lys Cys Ile Tyr Asp Ser Lys Pro Asp Phe Ser Pro
251     100                 105                 110
252 att gat taa ttgttttgta tttgactgaa ttttgacaat aaaggtacta      455
253 Ile Asp
254     115
256 tcgttatgaa aaaaaaaaaa aaaaaaa                               482
259 <210> SEQ ID NO: 7
260 <211> LENGTH: 133
261 <212> TYPE: PRT
262 <213> ORGANISM: Tenebrio molitor
264 <223> OTHER INFORMATION: Precursor Protein for Tm 12.84, Clones 2.2, 2.3, and 7.5
266 <400> SEQUENCE: 7
267 Met Lys Leu Leu Leu Cys Phe Ala Phe Ala Ala Ile Val Ile Gly Ala
268             -15                 -10                 -5
270 Gln Ala Leu Thr Asp Glu Gln Ile Gln Lys Arg Asn Lys Ile Ser Lys
271         1               5                   10
273 Glu Cys Gln Gln Val Ser Gly Val Ser Gln Glu Thr Ile Asp Lys Val
274 15                  20                  25                  30
276 Arg Thr Gly Val Leu Val Asp Asp Pro Lys Met Lys Lys His Val Leu
277                 35                  40                  45
279 Cys Phe Ser Lys Lys Thr Gly Val Ala Thr Glu Ala Gly Asp Thr Asn
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RAW SEQUENCE LISTING DATE: 08/15/2001
PATENT APPLICATION: US/09/876,348A TIME: 14:16:16

Input Set : A:\Rb125seq.txt
Output Set: N:\CRF3\08152001\I876348A.raw

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280             50                  55                  60
282 Val Glu Val Leu Lys Ala Lys Leu Lys His Val Ala Ser Asp Glu Glu
283         65                  70                  75
285 Val Asp Lys Ile Val Gln Lys Cys Val Val Lys Lys Ala Thr Pro Glu
286     80                  85                  90
288 Glu Thr Ala Tyr Asp Thr Phe Lys Cys Ile Tyr Asp Ser Lys Pro Asp
289 95                  100                 105                 110
291 Phe Ser Pro Ile Asp
292                 115
295 <210> SEQ ID NO: 8
296 <211> LENGTH: 115
297 <212> TYPE: PRT
298 <213> ORGANISM: Tenebrio molitor
300 <223> OTHER INFORMATION: Mature Protein for Tm 12.84, Clones 2.2, 2.3, and 7.5
302 <400> SEQUENCE: 8
303 Leu Thr Asp Glu Gln Ile Gln Lys Arg Asn Lys Ile Ser Lys Glu Cys
304 1               5                   10                  15
306 Gln Gln Val Ser Gly Val Ser Gln Glu Thr Ile Asp Lys Val Arg Thr
307             20                  25                  30
309 Gly Val Leu Val Asp Asp Pro Lys Met Lys Lys His Val Leu Cys Phe
310         35                  40                  45
312 Ser Lys Lys Thr Gly Val Ala Thr Glu Ala Gly Asp Thr Asn Val Glu
313     50                  55                  60
315 Val Leu Lys Ala Lys Leu Lys His Val Ala Ser Asp Glu Glu Val Asp
316 65                  70                  75                  80
318 Lys Ile Val Gln Lys Cys Val Val Lys Lys Ala Thr Pro Glu Glu Thr
319                 85                  90                  95
321 Ala Tyr Asp Thr Phe Lys Cys Ile Tyr Asp Ser Lys Pro Asp Phe Ser
322             100                 105                 110
324 Pro Ile Asp
325         115
328 <210> SEQ ID NO: 9
329 <211> LENGTH: 481
330 <212> TYPE: DNA
331 <213> ORGANISM: Tenebrio molitor
333 <223> OTHER INFORMATION: Non-His-tagged, Signal plus, Clone 3.4
335 <400> SEQUENCE: 9
336 ggcacgagca aaa atg aaa ctc ctc ttg tgc ttt gct ttc gcc gcc     46
337                Met Lys Leu Leu Leu Cys Phe Ala Phe Ala Ala
338                            -15                 -10
340 atc gtc atc gga gct cag gct ctc acc gac gaa cag ata cag aaa    91
341 Ile Val Ile Gly Ala Gln Ala Leu Thr Asp Glu Gln Ile Gln Lys
342         -5                  1                   5
344 agg aac aag atc agc aaa gaa tgc cag cag gtg tcc gga gtg tcc   136
345 Arg Asn Lys Ile Ser Lys Glu Cys Gln Gln Val Ser Gly Val Ser
346     10                  15                  20
348 caa gag acg atc gac aaa gtc cgc aca ggt gtc ttg gtc gac gat   181
349 Gln Glu Thr Ile Asp Lys Val Arg Thr Gly Val Leu Val Asp Asp
350     25                  30                  35
```

Please Note:
Use of n and/or Xaa have been detected in the Sequence Listing. Please review the Sequence Listing to ensure that a corresponding explanation is presented in the <220> to <223> fields of each sequence which presents at least one n or Xaa.

VERIFICATION SUMMARY DATE: 08/15/2001
PATENT APPLICATION: US/09/876,348A TIME: 14:16:17

Input Set : A:\Rbl25seq.txt
Output Set: N:\CRF3\08152001\I876348A.raw

L:11 M:271 C: Current Filing Date differs, Replaced Current Filing Date
L:1880 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:44
L:1888 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:44
L:1889 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:44
L:1912 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:44
L:1913 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:44
L:1916 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:44
L:1920 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:44
L:1934 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:45
L:1935 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:45
L:1938 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:45
L:1939 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:45
L:1942 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:45
L:1946 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:45
L:1950 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:45
L:1954 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:45
L:1958 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:45
L:1962 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:45
L:1966 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:45
L:1970 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:45
L:1974 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:45
L:1989 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:46
L:1993 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:46
L:1997 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:46
L:2001 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:46
L:2005 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:46
L:2009 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:46
L:2013 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:46
L:2017 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:46
L:2021 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:46
L:2025 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:46
L:2029 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:46
L:2044 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:47
L:2048 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:47
L:2052 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:47
L:2056 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:47
L:2060 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:47
L:2064 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:47
L:2068 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:47
L:2072 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:47
L:2076 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:47
L:2080 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:47
L:2084 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:47
L:2099 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:48
L:2102 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:48
L:2105 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:48
L:2108 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:48
L:2111 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:48

VERIFICATION SUMMARY DATE: 08/15/2001
PATENT APPLICATION: US/09/876,348A TIME: 14:16:17

Input Set : A:\Rb125seq.txt
Output Set: N:\CRF3\08152001\I876348A.raw

L:2114 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:48
L:2117 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:48
L:2120 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:48